

## CLAIMS

## 1. Decellularized tissue, wherein:

5       A) a cell survival rate of the tissue is less than a level at which an immune reaction is elicited in an organism; and

10      B) the tissue is not damaged to such an extent that hinders the tissue from exhibiting a function which was possessed by the tissue when the tissue was not decellularized.

2. Decellularized tissue according to claim 1, wherein the cell survival rate of the tissue is 30% or less.

15      3. Decellularized tissue according to claim 1, wherein the tissue damage rate of the tissue is 30% or less.

20      4. Decellularized tissue according to claim 1, wherein the tissue has a tissue strength which permits a clinical application.

25      5. Decellularized tissue according to claim 1, wherein the tissue has a tissue strength which is 80% or more of a tissue strength which was possessed by the tissue when the tissue was not decellularized.

30      6. Decellularized tissue according to claim 1, wherein the tissue has a tissue strength having a  $\beta$  value which is 80% or more of a  $\beta$  value which was possessed by the tissue when the tissue was not decellularized.

7. Decellularized tissue according to claim 1, wherein the tissue has a tissue strength having a  $\beta$  value of 20 or more.

8. Decellularized tissue according to claim 1, wherein the tissue is lumenal tissue.
- 5 9. Decellularized tissue according to claim 1, wherein the tissue is tissue selected from blood vessels, blood vessel-like tissue, cardiac valves, pericardia, dura mater, corneas, and bones.
- 10 10. Decellularized tissue according to claim 1, wherein a state of the tissue, in which the tissue is not damaged to such an extent that hinders the tissue from exhibiting a function which was possessed by the tissue when the tissue was not decellularized, includes that an extracellular matrix of the tissue is not substantially degenerated.
- 15 11. Decellularized tissue according to claim 10, wherein a survival rate of the extracellular matrix is at least about 50%.
- 20 12. Decellularized tissue according to claim 1, wherein the tissue is derived from a mammal.
- 25 13. Decellularized tissue according to claim 1, wherein the tissue is derived from a human.
14. Decellularized tissue according to claim 1, wherein the tissue is derived from a swine.
- 30 15. A tissue graft, comprising:
  - A) decellularized tissue, wherein a cell survival rate of the tissue is less than a level at which an immune reaction is elicited in an organism; the tissue is not damaged

to such an extent that hinders the tissue from exhibiting a function which was possessed by the tissue when the tissue was not decellularized; and the tissue has a desired structure.

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16. A tissue graft according to claim 15, wherein the tissue graft comprises a cell.

10 17. A tissue graft according to claim 16, wherein the cell is a recipient-derived cell.

18. A tissue graft according to claim 16, wherein the cell is derived from an organism of the same species as the tissue.

15 19. A tissue graft according to claim 15, wherein the tissue graft comprises no cell.

20 20. A tissue graft according to claim 15, wherein the decellularized tissue is tissue of an organ selected from the group consisting of blood vessels, blood vessel-like tissue, cardiac valves, pericardia, dura mater, corneas, and bones.

25 21. A tissue graft according to claim 15, wherein the tissue is derived from a mammal.

22. A tissue graft according to claim 15, wherein the tissue is derived from a human.

30 23. A tissue graft according to claim 15, wherein the cell is selected from the group consisting of vascular endothelial cells, smooth muscle cells, fibroblast, blood cells, and precursor cells and somatic stem cells capable of

differentiating thereto.

24. A membrane-like tissue graft, comprising:

5       A) decellularized tissue, wherein a cell survival  
rate of the tissue is less than a level at which an immune  
reaction is elicited in an organism; the tissue is not damaged  
to such an extent that hinders the tissue from exhibiting  
a function which was possessed by the tissue when the tissue  
was not decellularized; and the tissue has a desired  
10      structure.

25. A membrane-like tissue graft according to claim 24,  
wherein the tissue graft comprises a cell.

15      26. A membrane-like tissue graft according to claim 24,  
wherein the cell is a recipient-derived cell.

20      27. A membrane-like tissue graft according to claim 25,  
wherein the cell is derived from an organism of the same  
species as the tissue.

25      28. A membrane-like tissue graft according to claim 25,  
wherein the tissue graft comprises no cell.

30      30. A method of producing decellularized tissue, comprising  
the steps of:

- 1) providing tissue; and
- 2) immersing the tissue in a solution containing a  
non-micellar amphipathic molecule.

30      31. A method according to claim 30, further comprising:  
3) washing the tissue.

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32. A method according to claim 31, wherein the washing step is performed with PBS.

33. A method according to claim 30, wherein the amphipathic 5 molecule is a 1,2-epoxide polymer.

34. A method according to claim 30, wherein the amphipathic molecule is polyethylene glycol (PEG).

10 35. A method according to claim 34, wherein an average molecular weight of the PEG is between 200 to 6000.

36. A method according to claim 34, wherein an average molecular weight of the PEG is between 1000 and 2000.

15 37. A method according to claim 34, wherein an average molecular weight of the PEG is between 1500 and 2000.

20 38. A method according to claim 34, wherein an average molecular weight of the PEG is smaller than or equal to 1000.

39. A method according to claim 30, wherein the immersing step is performed for 30 min to 60 min.

25 40. A method according to claim 30, wherein the immersing step comprises physical treatment.

41. A method according to claim 30, wherein the washing step is performed for 3 days to 5 days.

30 42. A method according to claim 30, wherein the amphipathic molecule is biocompatible.

43. A method according to claim 30, wherein the tissue is tissue selected from the group consisting of blood vessels, blood vessel-like tissue, cardiac valves, pericardia, dura mater, corneas, and bones.

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44. A method according to claim 30, wherein the tissue is derived from a mammal.

10 45. A method according to claim 30, wherein the tissue is derived from a human.

46. A method according to claim 30, further comprising:

4) performing chemical treatment.

15 47. A method according to claim 31, further comprising:  
4) performing chemical treatment.

48. A method according to claim 46, wherein the chemical treatment is performed with DNase.

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49. A method according to claim 46, wherein the chemical treatment is performed with DNaseI.

25 50. A method according to claim 47, wherein the chemical treatment is performed with DNase.

51. A method according to claim 47, wherein the chemical treatment is performed with DNaseI.

30 52. A method according to claim 30, further comprising:  
disseminating a cell.

53. A decellularized tissue, obtained by a method according

to claim 30.

54. A tissue regeneration method, comprising the steps of:

5       a) providing decellularized tissue into an organism, wherein a cell survival rate of the tissue is less than a level at which an immune reaction is elicited in an organism; the tissue is not damaged to such an extent that hinders the tissue from exhibiting a function which was possessed by the tissue when the tissue was not decellularized; and

10      b) incubating the tissue within the organism for a time sufficient for the tissue to regenerate.

55. A method according to claim 54, further comprising providing a cell to the decellularized tissue.

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56. A method according to claim 55, wherein the cell is derived from the organism.

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57. A method according to claim 55, wherein the cell is present within the organism.

58. A method according to claim 55, wherein the cell is derived from a host homologous to the organism.

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59. A method according to claim 55, wherein the cell is derived from a host heterologous to the organism.

60. A method according to claim 55, wherein the cell is previously isolated from the organism.

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61. A method according to claim 55, wherein the cell is a blood vessel cell or a blood vessel-like cell.

62. A method according to claim 54, further comprising providing a physiologically active substance capable of inducing cell differentiation to the organism.

5 63. A method according to claim 62, wherein the physiologically active substance is derived from or foreign to the organism.

10 64. A method according to claim 62, wherein the physiologically active substance is provided in a form of a nucleic acid or a polypeptide.

15 65. A method according to claim 62, wherein the physiologically active substance is selected from the group consisting of HGF, VEGF, FGF, IGF, PDGF, and EGF.

20 66. A method according to claim 54, wherein the tissue is tissue selected from the group consisting of blood vessels, blood vessel-like tissue, cardiac valves, pericardia, dura mater, corneas, and bones.

67. A method of producing a tissue graft, comprising the steps of:

25 a) providing decellularized tissue into an organism, wherein a cell survival rate of the tissue is less than a level at which an immune reaction is elicited in an organism; the tissue is not damaged to such an extent that hinders the tissue from exhibiting a function which was possessed by the tissue when the tissue was not decellularized; and

30 b) causing a self cell in the organism to infiltrate the decellularized tissue; and

c) incubating the tissue within the organism for a time sufficient for the cell to differentiate.

68. A method according to claim 52, wherein the cell is a blood vessel cell or a blood vessel-like cell.

5 69. A method according to claim 67, wherein the tissue is tissue selected from the group consisting of blood vessels, blood vessel-like tissue, cardiac valves, pericardia, dura mater, corneas, and bones.

10 70. A method according to claim 67, wherein the decellularization tissue comprises a cell.

71. A method according to claim 67, wherein the decellularization tissue is autologous.

15 72. A method according to claim 67, wherein the decellularization tissue is derived from a homologous host.

73. A method according to claim 67, wherein the decellularization tissue is derived from a heterologous host.

20 74. A method according to claim 70, wherein the cell is autologous.

25 75. A method according to claim 70, wherein the cell is derived from a homologous host.

76. A method according to claim 70, wherein the cell is derived from a heterologous host.

30 77. A method according to claim 67, further comprising:  
d) providing a physiologically active substance capable of inducing differentiation of the cell.

78. A method according to claim 77, wherein the physiologically active substance is a cytokine having hematopoiesis activity.

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79. A tissue graft, produced by a method according to claim 67.

10 80. A method of treating a subject requiring implantation of tissue or an organ or treating a subject at a risk of implantation of tissue or an organ for prophylaxis, the method comprising the steps of:

15 1) providing decellularized tissue, wherein a cell survival rate of the tissue is less than a level at which an immune reaction is elicited in an organism; the tissue is not damaged to such an extent that hinders the tissue from exhibiting a function which was possessed by the tissue when the tissue was not decellularized, or a tissue graft comprising the decellularized tissue; and

20 2) implanting the decellularized tissue or tissue graft to the subject.

25 81. A method according to claim 80, wherein the tissue further comprises a cell.

82. A method according to claim 81, wherein the cell is a recipient-derived cell.

30 83. A method according to claim 81, wherein the cell is derived from an organism of the same species as the tissue.

84. A method according to claim 81, wherein the tissue comprises no cell.

85. A method according to claim 80, wherein the tissue is derived from the subject.

5 86. A method according to claim 80, wherein the tissue is tissue selected from the group consisting of blood vessels, blood vessel-like tissue, cardiac valves, pericardia, dura mater, corneas, and bones.

10 87. A method according to claim 80, wherein the subject is a mammal.

88. A method according to claim 80, wherein the subject is a human.

15 89. A medicament for organ implantation, comprising:  
A) decellularized tissue, wherein a cell survival rate of the tissue is less than a level at which an immune reaction is elicited in an organism; the tissue is not damaged to such an extent that hinders the tissue from exhibiting a function which was possessed by the tissue when the tissue was not decellularized, or a tissue graft comprising the decellularized tissue.

25 90. A medicament according to claim 89, wherein the tissue is tissue selected from the group consisting of blood vessels, blood vessel-like tissue, cardiac valves, pericardia, dura mater, corneas, and bones.

30 91. A medicament according to claim 89, wherein the cell is derived from a mammal.

92. A medicament according to claim 89, wherein the tissue

is derived from a human.

93. A medicament according to claim 89, wherein the tissue  
is derived from the subject requiring implantation.

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94. A medicament according to claim 89, wherein the tissue  
is derived from a swine.

95. Use of decellularized tissue, wherein a cell survival  
10 rate of the tissue is less than a level at which an immune  
reaction is elicited in an organism; the tissue is not damaged  
to such an extent that hinders the tissue from exhibiting  
a function which was possessed by the tissue when the tissue  
was not decellularized, or a tissue graft comprising the  
15 decellularized tissue, for producing a medicament for organ  
implantation.